

**SPECIFICATION AMENDMENTS**

**Page 2, lines 7-10**, within the paragraph of these pages and line numbers, please enter the following amendments:

Microfluidic capillary array electrokinetic (CAEK) devices are provided employing individual units having four fold symmetry, each unit providing two four separate subunits permitting two independent determinations, where four subunits of ~~different units~~ share a single supply reservoir for a total of 8 determinations.

**Page 3, lines 3-12**, within the paragraph of these pages and line numbers, please enter the following amendments:

Each unit is characterized by having four-fold symmetry, where each unit may be divided further into ~~half-units~~ quarter-units or subunits having two single assay units to provide a total of 8-fold symmetry in relation to a common supply reservoir. ~~Each unit in each of the designs comprises four assay units.~~ Each unit has a central waste supply reservoir common to all of the assay units and at least one more waste reservoir shared with other units. The assay units are characterized by having a reagent source, which meets at an intersedction, usually a T, with a compound source, frequently a test or candidate compound or a labeled reagent, and connects to a delivery channel, where unused compound and reagent are directed to a common waste reservoir. The reagent source provides reagent to 8 single assay units, i.e. four ~~half-units~~ quarter-units, where the reagent is distributed to the 2 single assay units in each ~~half-unit~~ quarter-unit.

**Page 3, line 27**, within the paragraph of these pages and line numbers, please enter the following amendments:

The crosses between the assay channel and the delivery channel are on opposite sides of a buffer reservoir. The design allows for a different composition for each of the assay units, while permitting common use of reagent, buffer and waste reservoirs. The design provides that the different components used in the assay move to a waste reservoir common to four assay units, that the assay mixture may be injected into the assay channel from the delivery channel and that a common buffer reservoir provides a continuous source of liquid for transport of the assay

mixture downstream toward a second common ~~waster~~ waste reservoir past a detector. The assay units will usually be associated with a single sample or test compound, with one or more assay units associated with a control.

**Page 4, lines 17 – 20**, within the paragraph of these pages and line numbers, please enter the following amendments:

The disposition of the units is to have ~~half units extending the full length along two edges and full units between the half units and at the other two edges. In this way, each column is bordered by half units and the first and last lines have half units, while the remaining lines have full units~~ adjacent units aligned so as to share components of similar function. In this way, multiple units can be arranged in a device substrate that makes the most efficient use of space.

**Page 5, line 11 and line 14**, within the paragraph of these pages and line numbers, please enter the following amendments:

The total surface area will generally be in the range of about 9 to 200cm<sup>2</sup>, more usually about 12 to 150cm<sup>2</sup>. Particularly, the total surface area occupied by 24 12 units will usually conform to a 96 microtiter well plate, generally being 8 by 12 cm, with 9mm spacings. By contrast where one wishes to have a 384 assay format, 96 units generally conform to a 384 ~~meirotiter~~ microtiter well plate, being 8 by 12cm, with 4.5mm spacings.. The dimensions of the channels and reservoirs will vary with the size of the units, where the dimensions will generally be larger, the larger the size of the unit. The volume of the reservoirs will vary depending on their function, the reagent reservoirs generally being in the range of 100nl to 1µl while the buffer and waste reservoirs will generally have a volume in the range of about 1 to 10µl.

**Page 7, line 10, line 14, line 15 and line 16**, within the paragraph of these pages and line numbers, please enter the following.

For further understanding of the invention, the drawings will now be considered. In Fig. 1a is depicted a plan view of a substrate design for a an assay unit for a 96 assay format. The purpose of the assay unit is to detect the interaction between a test compound and a target

compound, e.g. an enzyme. The design 100 comprises a reagent reservoir 102 connected to delivery channel 104. Joined to delivery channel 104 is test compound reservoir 106 through side channel 108, which channels join at T ~~junction~~ junction 110. The delivery channel 104 includes an incubation region or channel 105 and connects to waste reservoir 112 crossing assay channel 114 at cross-juncture 116. As indicated previously, the cross-~~junction~~ junction may be a cross where the crossing channels stay in the same line or channels on opposite sides of a straight channel may be displaced so as to form a double-T intersection, where the spacing between the two channels serves to define the volume that is injected into the straight channel, in this case the assay channel 114. Assay channel 114 connects buffer reservoir 118 to waste reservoir 120. Electrodes (not shown) are present in all of the reservoirs. Upstream from waste reservoir 120 is detector 122.

**Page 8, lines 4 – 13**, within the paragraph of these pages and line numbers, please enter the following amendments:

Initially, electrodes in reagent reservoir 102, test compound and substrate reservoir 106 and waste reservoir 112 are activated to provide a field, which moves the enzyme, test compound and substrate into delivery channel 104 for incubation in incubation channel 105 ~~where the mixture moves toward T junction 110 so that the incubated mixtures arrives at the T junction. Further reaction may occur as the mixture is injected into the assay channel 114.~~ When the components reach T junction 110, the components mix to form the assay mixture and the enzyme reacts with the substrate in relation to the effect of the test compound. The amount of enzyme product produced is related to the activity of the test compound. The field may be maintained while the enzyme is moving toward cross-juncture 116 and enzyme product is continuously being produced. Further reaction may occur as the mixture is injected into the assay channel 114.

**Page 8, line 19 – page 9, line 4**, within the paragraph of these pages and line numbers, please enter the following amendments:

In Fig. 1b is shown a device 150 having substrate 152. A pattern of units 154 ~~and half units 156~~ are shown, which is referred to as an 8-plex on a 96-assay format. The units 154 are repeated across ~~Two two~~ rows 158 and 160 of half units 156 border the units 154 along two edges and six columns. The units 154 are ~~four-eight~~ units of 100 organized so as to share the

maximum number of channels and reservoirs compatible with the purpose for which the device is used. In unit 154 there are ~~four~~ eight test compound and substrate reservoirs ~~106b1-4~~ 106a-h, associated with ~~individual single~~ assay units ~~100b1-4~~ 100, as shown in Fig. 1a. The reagent reservoir ~~102b~~ 102a supplies the reagent to eight assay units ~~100b~~ 100, where the eight assay units are ~~divided into~~ provided in four ~~half-units~~ quarter-units 156, as part of four units 154. Each ~~half-unit~~ quarter-unit 156, individually or as part of ~~individual unit~~ 156 154, has a common buffer reservoir ~~118b~~ 118a-d. There are ~~two~~ six waste reservoirs ~~120b~~ 120a-f associated with each unit 154 and outside the design pattern for the unit, where the waste from the assay ~~channel~~ 114b channels 114a-h is directed. A single waste reservoir ~~112b~~ , e.g. 112a, for the delivery channel is central to the pattern of the ~~unit~~ 154 subunit 156, receiving the waste from ~~all four the~~ two delivery channels. In this way for ~~four assay units~~ 100b each subunit 156, there are a total of three waste reservoirs, two reservoirs ~~120b~~ 120 for the ~~four assay channels~~ 114b and one waste reservoir ~~112b~~ 112 for the ~~four delivery channels~~ 104b available to be shared with an adjacent unit. In addition, there is one reagent reservoir ~~102b~~ for eight assay units ~~100b~~. Detection sites are closely confined and symmetrical to permit a single detection unit, such as a CCD to be employed, or be able to move detection systems along a line or row for determinations.

Page 9, line 9 and line 12, within the paragraph of these pages and line numbers, please enter the following amendment:

Figs. 2a, 2b and 2c depict a device 200 employing a similar unit design as in Fig. 1a, which is organized as 8-plex symmetry for a 384-assay format. The device is broken down into a single assay unit or unit cell in Fig. 2b and a unit with 8-plex symmetry in Fig. 2c. Selecting one unit 202, the unit has a reagent reservoir 204, which supplies reagent through reagent supply channel 206. There are ~~four~~ eight test compound reservoirs 208 symmetrically positioned. Test compound reservoir is connected to delivery channel 210 through side channel 212. Delivery channel 210 is connected to common waste reservoir 214. Delivery channel 210 connects with assay channel 216 at cross-junction 218. Assay channel ~~220~~ 216 connects buffer reservoir 222 with waste reservoir 224.

**Page 10, line 4**, within the paragraph of these pages and line numbers, please enter the following amendment:

As shown in Fig, 3a, the network assay unit 300, has a reactor reservoir 302, which for the purposes of the present illustration is a PCR reactor. The reactor is equipped for thermal cycling (not shown). The sample DNA and reagents, namely DNA polymerase, probes and dNTPs, in an appropriate buffer, are introduced into reactor reservoir 302, and a number of thermal cycles performed resulting in the amplification of target DNA. The PCR reactor 302 is connected through delivery channel 304 and through side channel 306 to capture bead reservoir 308 and through delivery channel 304 and side channel 310 to buffer reservoir 312. Delivery channel 304 continues in a tortuous route, where labeled probe reservoir 314 is connected through side channel 316 to delivery channel 304. After the connection with side channel 316 delivery channel 304 has a bead trap region 318. The delivery channel 304 continues through bead trap region 318 to waste reservoir 320. Delivery channel 304 crosses assay channel 322 at a cross-junction 324. Assay channel 322 connects buffer reservoir 326 to waste reservoir 328. The assay channel 322 passes detector 330 upstream from waste reservoir 328. Electrodes (not shown) are present in the different reservoirs.

**Page 11, line 19**, within the paragraph of these pages and line numbers, please enter the following amendments:

Once the labeled probe is released from the beads, it is then moved from the bead trap region 318 by means of the electrical field to the cross-intersection 324 between the delivery channel 304 and the assay channel 324 322. By changing the electrical field from across the delivery channel 304 to across the assay channel 322, the slug at the intersection 324 containing the labeled probes can be transported into the assay channel 322 toward the detection system, the buffer reservoir 326 providing the fluid for the movement in the assay channel 322. When the labeled probes move past the detector 330, the label may be detected, indicating the presence of a particular sequence in the DNA sample. Depending on the purpose of the assay and the labeled probes, the method may be multiplexed, so that a number of differently labeled probes, which can be independently detected in relation to a particular sequence, may be detected to define specific sequences in the sample DNA.

**Page 11, line 28 – page 12, line 10**, within the paragraph of these pages and line numbers, please enter the following amendments:

In Fig. 3b, a unit consisting of ~~four~~ eight assay units depicted in Fig. 3a is shown. The unit 350b employs a reagent constituent comprising a reagent reactor 302b, particularly in the present illustration, a PCR reactor, a capture bead reservoir 308b, a buffer reservoir 312b, connected together through delivery channel 304b and side channels 306b and 310b. The delivery channel 304b feeds the amplified DNA from the PCR reactor 302b partially bound to the beads from bead reservoir 308b to eight different assay units ~~300b~~ 300, as shown in Fig. 3a ~~present as four half-units 352b~~. Considering only one of the assay units in view of the symmetry of the system, labeled probe reservoir 314b feeds labeled probe through side channel 316b into delivery channel 304b to bind to DNA captured by the beads from bead reservoir 308b. The beads with the sample DNA and labeled probe, if the assay is positive, are captured by the bead trap 318b. The labeled probe is then released from the beads and transported to the delivery channel 304b and assay channel 322b cross-intersection 324b. The labeled probe is injected into the assay channel 322b by means of buffer from buffer reservoir 326b and the electrical field provided by electrodes in buffer reservoir 326b and waste reservoir 328b. A detector ~~330b~~ detects the passage of the labeled probe through the assay channel 322b.